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(54) Title: NOVEL STABILIZED CAROTENOID COMPOSITIONS

(57) Abstract: Compositions comprising a carotenoid and/or fat-soluble vitamin in a matrix of a protein which is cross-linked with a reducing substance, a solid vegetable fat and, optionally, auxiliary agents provide increased stability of the carotene and/or vitamin when incorporated into food, particularly a vegetable fat.

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Novel stabilized carotenoid compositions

The present invention relates to novel stabilized carotenoid compositions. More particularly, the present invention relates to compositions comprising a carotenoid and/or fat-soluble vitamin in a matrix of a protein which is cross-linked with a reducing substance, a solid vegetable fat and, optionally, auxiliary agents.

- 10 The invention further relates to a process for the preparation of the novel compositions; to the use of these compositions as additives, particularly as functional health ingredients for food, particularly vegetable fats; and to food, particularly vegetable fats containing a novel composition of this invention.

The term "functional health ingredient" refers to additives for food which supplement or  
15 improve the nutritional value of food, such as vitaminizing compositions.

The term "carotenoid" as used herein comprises a natural or synthetic carotene or structurally related polyene compound which can be used as a functional health ingredient or colorant for food, such as  $\alpha$ - or  $\beta$ -carotene, 8'-apo- $\beta$ -carotenal, 8'-apo- $\beta$ -carotenoic acid esters such as the ethyl ester, canthaxanthin, astaxanthin, lycopene, lutein, zeaxanthin  
20 or crocetin, or mixtures thereof. The preferred carotenoids are  $\beta$ -carotene, lycopene and lutein and mixtures thereof, especially  $\beta$ -carotene. The amount of carotenoid and/or fat-soluble vitamin in the compositions of the present invention may be from about 0.1 to about 30 wt.-%, and preferably is from about 1 to about 10 wt.-%.

- 25 The protein is preferably gelatin, which may be bovine, swine or fish gelatin or hydrolyzed gelatin. Any gelatin which has a bloom number in the range of practically 0 to about 300, particularly of about 50 to about 250 can be employed in the practice of this invention. Both Type A and Type B can be employed. The amount of protein in the compositions of

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the present invention may be from about 5 to about 80 wt.-%, and preferably is from about 10 to about 40 wt.-%.

5 The reducing substance is preferably a reducing sugar such as fructose, glucose, lactose, maltose, xylose, arabinose, ribose, invert sugar or high fructose or glucose syrups. Also, aldehydes such as glutar aldehyde may be used as the reducing substance.

The amount of reducing substances in the compositions of the present invention may be from about 5 to about 50 wt.-%, and preferably is from about 10 to about 35 wt.-%.

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The vegetable fat should be solid at room temperature (20-25 °C). Examples of such fats are plant fats and hardened (i.e. hydrogenated) plant oils such as hardened sunflower oil (also referred to as sunflower fat) and hardened rape oil. Further examples of vegetable fats for use in the present invention are hardened ricinus oil, hardened cotton seed oil, cocos  
15 fat and palm fat.

The amount of solid vegetable fat in the compositions of the present invention may be from about 2 to about 30 wt.-%, and preferably is from about 5 to about 20 wt.-%.

20 Fat-soluble vitamins which may be present in the compositions of the present invention are vitamin A, D, E and K. These vitamins may be present singly or in any desired combination, or in combination with a carotenoid as defined above. In a preferred aspect the present invention relates to compositions comprising a carotenoid in a matrix of a protein which is cross-linked with a reducing substance, a solid vegetable fat and, optionally, auxiliary agents.

25 The compositions of the present invention may include further ingredients, e.g. auxiliary agents such as anti-oxidants, emulsifiers, humectants, extenders, solubilizers, and other coloring agents; or nutritionally valuable agents, e.g., water-soluble vitamins such vitamin B<sub>1</sub>, B<sub>2</sub>, B<sub>6</sub> or C. Examples of antioxidants are butylated hydroxyanisol, butylated hydroxytoluene and ethoxyquin. The emulsifier may be a lecithin. Examples of humectants  
30 are glycerol, sorbitol, propylene glycol and polyethylene glycols. Preferably, the compositions of the present invention contain glycerol in an amount of e.g. 5 wt.-% to about 30 wt.-%.

In accordance with the present invention the novel compositions can be prepared by forming an emulsion of an aqueous solution of the protein, the reducing substance and

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optional water-soluble auxiliary agents with the carotenoid and/or fat-soluble vitamin, the solid vegetable fat and optional fat-soluble auxiliary agents, converting the emulsion into a dry particulate form and submitting the particles to conditions to effect cross-linking of the protein and the reducing substance.

- 5 In one embodiment of the process of this invention the protein, e.g., gelatin is dissolved in water, if necessary, by gentle warming and the carotenoid and other fat-soluble ingredients are the dispersed or emulsified in the solution of the protein. The carotenoid and other fat-soluble ingredients may be added as such or dissolved in an appropriate organic solvent, e.g., a chlorinated hydrocarbon such as chloroform. The reducing substance and optional
- 10 water-soluble ingredients can be introduced into the mixture either before or after adding the carotenoid and other optional fat-soluble ingredients. The mixture is homogenized by conventional techniques such as agitating, high-pressure homogenisation, high-shear emulsification or the like and the resulting emulsion converted into a dry particulate form such as granules or beadlets, by spraying onto a bed of starch. If a solvent has been used to
- 15 dissolve the carotenoid or optional fat-soluble agents such solvent is suitably be removed from the emulsion by evaporation prior to spray-drying.

- The starch used in the process to collect the sprayed emulsion may consist entirely of a powder of starch and/or chemically modified starch. The starch may also contain minor amounts of lubricants or other modifiers such as talc, silicic acid, flours, hydrogenated fats
- 20 and metal salts of higher fatty acids, e.g., calcium stearate. The starch powder should be substantially insoluble in cold water and be resistant to wetting by water; it should have an appreciable capacity to absorb and/or adsorb water; and it should be free-flowing. Its moisture content should be below about 10 wt.-%. Starch powders of the desired type are e.g. those disclosed in US 2,613,206 and those commercially available as "Dry-Flo" from
- 25 National Starch Products, Inc., New York. The spraying of the emulsion obtained in accordance with the process of this invention may be carried out by techniques known per se, e.g. as disclosed in US 4,670,247 the contents of which are enclosed herewith for reference purposes. Most preferred is spray-drying in combination with fluidized-bed granulation (commonly known as fluidized spray drying or FSD). The spraying conditions
- 30 are suitably adjusted (by the size of the spraying nozzle orifice, the percentage of water in the emulsion etc.) so as to produce particles passing a 10 mesh screen and being retained by a 200 mesh screen, preferably particles of a size in the range between 20 and 170 mesh.

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The granules or beadlets obtained are then submitted to conditions to effect cross-linking of the protein, e.g., gelatin, and the reducing substance (sugar). The cross-linking may be accomplished by heat-treatment or by treatment with enzymes, e.g., transglutaminase.

Preferably, the cross-linking is accomplished by heat-treatment. In one embodiment of  
5 the process of the present invention, the granules or beadlets are heated to about 60 °C to about 100 °C for about 10 to about 60 minutes, e.g. to 80 °C for about 40 minutes. The so-obtained composition represents a powder which is insoluble in boiling water.

The compositions of the present invention may find use particularly as additives to vegetable fats such as margarine where they provide increased stability of the particular  
10 ingredient as compared to similar compositions which do not contain a hardened vegetable fat. They may be present, e.g., in vegetable fats in an amount to provide from about 1 to about 2000 ppm of carotenoid.

The following Example illustrates the invention further.

Example

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44 g of gelatin (Bloom 140), 48 g of fructose, 20 g of glycerol and 3 g of ascorbyl palmitate were dissolved in water at 60 °C. The pH was adjusted to 7.6 by the addition of 2.4 ml of 20 % (wt/vol) aqueous sodium hydroxide solution to yield Phase I. 11 g of lutein cake base (75 %, Kemin Foods, Des Moines, Iowa, USA), 4.8 g of lycopene (70%, LycoRed Natural  
20 Products Industries Ltd., Israel), 20 g of sunflower fat and 3 g of d,l- $\alpha$ -tocopherol were dissolved in 150 ml of chloroform with heating to reflux. There was thus obtained phase II. Phase II was slowly emulsified into Phase I at 45 °C using a rotor-stator emulsifier to obtain a particle size of the inner phase of 214 nm. The chloroform was distilled off and the resulting emulsion sprayed into a fluidized starch bed. The so-obtained beadlets were  
25 separated by sieving and dried in a fluidized bed. The so-obtained a powder was transferred to a round-bottom flask and heated for 40 minutes in an oil bath of 80 °C in a rotary dryer.

The so-obtained product was incorporated into margarine in carotenoid concentrations of 260 ppm and 2000 ppm. After storage for four weeks at 4 °C the retention of the  
30 carotenoids was determined by spectrophotometry and was found to be 100 %.

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What is claimed is:

1. Compositions comprising a carotenoid and/or fat-soluble vitamin in a matrix of a protein which is cross-linked with a reducing substance, a solid vegetable fat and, optionally, auxiliary agents.
- 5 2. Compositions comprising a carotenoid in a matrix of a protein which is cross-linked with a reducing substance, a solid vegetable fat and, optionally, auxiliary agents.
3. Compositions as in claim 1 or 2 wherein the protein is gelatin.
4. Compositions as in any one of claims 1-3 wherein the reducing substance is a reducing  
10 sugar.
5. Compositions as in any one of claims 1-4 wherein the matrix comprises about 5 to about 80 wt.-% of protein, about 5 to about 50 wt.-% of a reducing substance, and about 2 to about 30 wt.-% of a solid vegetable fat.
6. Compositions as in any one of claims 1-5 wherein the matrix comprises about 10 to  
15 about 40 wt.-% of protein, about 10 to about 35 wt.-% of a reducing substance, and about 5 to about 20 wt.-% of a solid vegetable fat.
7. Compositions as in any one of claims 1-6 comprising about 0.1 to about 30 wt.-% of a carotenoid and/or fat-soluble vitamin.
8. Compositions as in any one of claims 1-6 comprising about 1 to about 10 wt.-% of a  
20 carotenoid and/or fat-soluble vitamin.
9. Compositions as in any one of claims 1-8 wherein the carotenoid  $\alpha$ - or  $\beta$ -carotene, 8'-apo- $\beta$ -carotenal, 8'-apo- $\beta$ -carotenoic acid esters such as the ethyl ester, canthaxanthin, astaxanthin, lycopene, lutein, zeaxanthin or crocetin, or mixtures thereof.
10. Compositions as in any one of claims 1-9 wherein the carotenoid  $\beta$ -carotene, lutein or  
25 lycopene, or mixtures thereof
11. Compositions as in any one of claims 1-10 wherein the carotenoid is  $\beta$ -carotene.
12. Compositions as in any one of claims 1-10 wherein the carotenoid is a mixture of lycopene and lutein.

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13. Compositions as in any one of claims 1-12 wherein the vegetable fat is sunflower fat or hardened rape oil.
14. Compositions as in any one of claims 1-12 wherein glycerol is present as an auxiliary agent.
- 5 15. Compositions as in claim 1 wherein the fat-soluble vitamin is vitamin A, D, E or K or mixtures thereof.
- 10 16. Process for preparing a composition as claimed in any one of claims 1-15 which comprises forming an emulsion of an aqueous solution of the protein, the reducing substance and optional water-soluble auxiliary agents with the carotenoid and/or fat-soluble vitamin, the solid vegetable fat and optional fat-soluble auxiliary agents, converting the emulsion into a dry particulate form and submitting the particles to conditions to effect cross-linking of the protein and the reducing substance.
17. The use of a composition as claimed in any one of claims 1-15 as an additive for food.
18. The use as in claim 17 wherein the food is a vegetable fat..
- 15 19. The use as in claim 17 wherein the food is margarine.
20. Food containing a composition as claimed in any one of claims 1-15.
21. A vegetable fat containing a composition as claimed in any one of claims 1-15.
22. Margarine containing a composition as claimed in any one of claims 1-15.
23. The invention as described hereinbefore, particularly with reference to the Example.

# INTERNATIONAL SEARCH REPORT

International Application No  
PCT/EP 02/09096

## A. CLASSIFICATION OF SUBJECT MATTER

IPC 7 A23L1/303 A23L1/305 A23L1/275

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 7 A23L

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the International search (name of data base and, where practical, search terms used)

EPO-Internal, WPI Data, PAJ, FSTA

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	US 5 356 636 A (SCHNEIDER JOACHIM U ET AL) 18 October 1994 (1994-10-18) column 1, line 1 - line 30 column 3, paragraphs 2,4,5 column 4, paragraphs 1,2,4,10 column 5, paragraph 2 column 7-8, table 1, examples 5-7	1-23
Y	EP 0 410 236 B (HOFFMANN LA ROCHE) 30 January 1991 (1991-01-30) page 2, paragraph 1 - paragraph 3 page 2, line 57 - page 3, line 42 page 4; examples 1,2	1-23
A	US 5 126 328 A (BOWER DAVID K ET AL) 30 June 1992 (1992-06-30) the whole document	1-23
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☒ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

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## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
L	<p>CORTESI R ET AL: "Sugar cross-linked gelatin for controlled release: microspheres and disks" BIOMATERIALS, ELSEVIER SCIENCE PUBLISHERS BV., BARKING, GB, vol. 19, no. 18, September 1998 (1998-09), pages 1641-1649, XP004161435 ISSN: 0142-9612 the whole document</p>	1-23

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Information on patent family members

International Application No

PCT/EP 02/09096

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